Defined culture of human embryonic stem cells and xeno-free derivation of retinal pigmented epithelial cells on a novel, synthetic substrate.

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Public Summary:
Age-related macular degeneration (AMD), a leading cause of blindness, is characterized by the death of the retinal pigmented epithelium (RPE), which is a monolayer posterior to the retina that supports the photoreceptors. Human embryonic stem cells (hESCs) can generate an unlimited source of RPE for cellular therapies, and clinical trials have been initiated. However, protocols for RPE derivation using defined conditions free of nonhuman derivatives (xeno-free) are preferred for clinical translation. This avoids exposing AMD patients to animal-derived products, which could incite an immune response. In this study, we investigated the maintenance of hESCs and their differentiation into RPE using Synthemax II-SC, which is a novel, synthetic animal-derived component-free, RGD peptide-containing copolymer compliant with good manufacturing practices designed for xeno-free stem cell culture. Cells on Synthemax II-SC were compared with cultures grown with xenogeneic and xeno-free control substrates. This report demonstrates that Synthemax II-SC supports long-term culture of H9 and H14 hESC lines and permits efficient differentiation of hESCs into functional RPE. Expression of RPE-specific markers was assessed by flow cytometry, quantitative polymerase chain reaction, and immunocytochemistry, and RPE function was determined by phagocytosis of rod outer segments and secretion of pigment epithelium-derived factor. Both hESCs and hESC-RPE maintained normal karyotypes after long-term culture on Synthemax II-SC. Furthermore, RPE generated on Synthemax II-SC are functional when seeded onto parylene-C scaffolds designed for clinical use. These experiments suggest that Synthemax II-SC is a suitable, defined substrate for hESC culture and the xeno-free derivation of RPE for cellular therapies.

Scientific Abstract:
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